



Hydrolysed formula and risk of allergic or autoimmune disease: systematic review and meta-analysis

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ABSTRACT OBJECTIVE

To determine whether feeding infants with hydrolysed formula reduces their risk of allergic or autoimmune disease.

DESIGN

Systematic review and meta-analysis, as part of a series of systematic reviews commissioned by the UK Food Standards Agency to inform guidelines on infant feeding. Two authors selected studies by consensus, independently extracted data, and assessed the quality of included studies using the Cochrane risk of bias tool.

DATA SOURCES

Medline, Embase, Web of Science, CENTRAL, and LILACS searched between January 1946 and April 2015.

ELIGIBILITY CRITERIA FOR SELECTING STUDIES

Prospective intervention trials of hydrolysed cows' milk formula compared with another hydrolysed formula, human breast milk, or a standard cows' milk formula, which reported on allergic or autoimmune disease or allergic sensitisation.

RESULTS

37 eligible intervention trials of hydrolysed formula were identified, including over 19 000 participants. There was evidence of conflict of interest and high or unclear risk of bias in most studies of allergic outcomes and evidence of publication bias for studies of eczema and wheeze. Overall there was no consistent evidence that partially or extensively hydrolysed formulas reduce risk of allergic or autoimmune outcomes in infants at high pre-existing risk of these outcomes. Odds ratios for eczema at age 0-4, compared with standard cows' milk formula, were 0.84 (95% confidence interval 0.67 to 1.07; $I^2=30\%$) for partially hydrolysed formula; 0.55 (0.28 to 1.09; $I^2=74\%$) for extensively hydrolysed casein based formula; and 1.12 (0.88 to 1.42; $I^2=0\%$) for extensively

hydrolysed whey based formula. There was no evidence to support the health claim approved by the US Food and Drug Administration that a partially hydrolysed formula could reduce the risk of eczema nor the conclusion of the Cochrane review that hydrolysed formula could prevent allergy to cows' milk.

CONCLUSION

These findings do not support current guidelines that recommend the use of hydrolysed formula to prevent allergic disease in high risk infants.

REVIEW REGISTRATION

PROSPERO CRD42013004252.

Introduction

Immune mediated health conditions such as allergic and autoimmune diseases seem to have increased in prevalence in many countries and are leading causes of chronic illness in young people.¹ There is evidence that dietary exposures in infancy can influence the risk of these diseases, with a specific concern that early exposure to intact cows' milk protein in the form of infant formula could trigger the onset of allergic or autoimmune disease.²⁻⁴ Current infant feeding guidelines in North America, Australasia, and Europe recommend the use of hydrolysed formula in the first 4-6 months of life in place of a standard cows' milk formula for the primary prevention of allergic diseases in childhood.⁵⁻⁷ This has also been supported by the US Food and Drug Administration (FDA)⁸ and by a Cochrane systematic review.^{9,10} To inform guidelines on infant feeding in the United Kingdom, we undertook a systematic review of hydrolysed formula for preventing allergic sensitisation, allergic disease, or autoimmune disease.

Methods

This review is reported in accordance with PRISMA guidance. The review is one of a series of systematic reviews commissioned by the UK Food Standards Agency to inform UK guidelines on infant feeding, under the title "review of scientific published literature on infant feeding and development of atopic and autoimmune disease." The protocols for the systematic reviews were registered with the International Prospective Register of Systematic Reviews (PROSPERO CRD42013003802 "milk feeding"; CRD42013004239 "timing of allergenic food introduction"; CRD42013004252 "maternal and infant diet") on 5 August 2013, before titles were screened or studies selected from the search results. This review of hydrolysed formula is part of CRD42013004252 "maternal

WHAT IS ALREADY KNOWN ON THIS TOPIC

Breastfeeding is the optimum mode of nutrition for infants

Substitution with infant formula has been associated with allergic and autoimmune disease

International guidelines recommend use of a hydrolysed formula in place of standard infant formula for infants at risk of allergic disease to prevent eczema and allergy to cows' milk

WHAT THIS STUDY ADDS

There is no consistent evidence to support the use of hydrolysed formula for the prevention of allergic or autoimmune disease

and infant diet.” As part of this project we also searched for other systematic reviews covering the same topic published since 1 January 2011 with a revised AMSTAR score ≥ 32 .¹¹ No such reviews were identified for hydrolysed formula.

Study interventions and comparators

We included studies of any hydrolysed formula of cows' milk origin as the intervention of interest, compared with any non-hydrolysed cows' milk formula, human milk, or another type of hydrolysed cows' milk formula. Also included were studies in which hydrolysed formula was given as part of a multifaceted intervention, which we defined as an intervention with at least two other components in addition to the hydrolysed formula—for example, exclusion of allergenic food from the mother's diet, promotion of breastfeeding, delayed introduction of solid food, or measures to avoid exposure to house dust mite. We also included studies in which other interventions were applied to both intervention and control groups, such as exclusion of cows' milk from the mother's diet during lactation. Studies of hydrolysed formula of milk other than cows' milk, such as hydrolysed rice, goats' milk, or soya formula, were not included. We did not use a specific definition of partially or extensively hydrolysed formula but relied on the definition and/or trade name of formula as stated in the individual studies.

Study designs and populations

We included all intervention trials and searched for observational studies but did not extract data from these because of the evidence from intervention trials. Intervention trials were classified as randomised controlled trials, in which the method of treatment allocation was random; as quasi-randomised controlled trials, in which the method of treatment allocation was not totally random (but was thought unlikely to lead to imbalance between treatment groups in variables relevant to the outcome measures); and as controlled clinical trials, in which treatment allocation was non-random and likely to lead to imbalance between treatment groups. Controlled clinical trials were analysed separately from randomised controlled trials/quasi randomised controlled trials. We included studies of infant feeding between birth and 12 months of age and excluded studies in which infants or their mothers were defined by the presence of a pre-existing disease state, including very low birth weight or premature infants.

Study outcomes

Atopic and autoimmune outcomes were selected on the basis of their population prevalence in children and young adults in the UK and/or other affluent nations. We included diseases with a prevalence of at least one in 1000 children/adolescents or young adults (aged <40) but did not include rarer diseases. Atopic outcomes included were asthma (categorised as wheeze, recurrent wheeze, atopic wheeze, bronchial hyper-reactivity, forced vital capacity, peak

expiratory flow rate, forced expiratory volume in one second), eczema, allergic rhinitis and/or conjunctivitis, food allergy, allergic sensitisation (that is, skin prick or specific immunoglobulin E (sIgE) assessment, or total IgE level). Autoimmune outcomes included were type 1 diabetes mellitus (defined serologically and/or clinically), coeliac disease (defined serologically and/or clinically), inflammatory bowel disease, autoimmune thyroid disease, juvenile rheumatoid arthritis, vitiligo, or psoriasis. For atopic outcomes, we grouped age at assessment as 0-4, 5-14, 15-24, 25-44, 45-64, and ≥ 65 . Because of a paucity of outcome data in adults, we pooled age groups ≥ 15 . Autoimmune outcomes were not stratified for analyses by age at outcome assessment.

Data sources

The search strategies included both text terms and subject heading terms where appropriate and were initially developed for Medline and then adapted for other databases. We searched the Cochrane Library (2013, issue 7), Embase (1947 to July 2013), LILACS (1982 to July 2013), Medline (1946 to July 2013), and Web of Science (1970 to July 2013) on 25 July 2013 and reran the searches on 17 April 2015. We included all studies published up to that date and studies in progress or completed but unpublished identified through <http://apps.who.int/trialsearch/>. Peer reviewed publications and abstract publications were included if they contained data that had not subsequently been published as a peer reviewed publication. We reviewed the bibliography of eligible studies for possible additional publications and included all eligible publications, regardless of the language. Authors of eligible or potentially eligible studies were not contacted for original data. We extensively piloted and refined the search strategies to optimise sensitivity, comparing search results with those of other published systematic reviews. Appendix 1 shows the Medline search strategies for the complete systematic review project.

Study selection

Seven trained researchers (RJB, VG-L, DI, NG, KJ, JC, ZR) screened titles and abstracts in duplicate. Two researchers screened titles independently and met to agree on inclusion. Their screening was checked by a third member of the team, and uncertainties were brought to a full team meeting for discussion. This procedure took place between February and April 2014 and in April and May 2015 (RJB, TK, VG-L), with weekly team meetings to discuss uncertainties about study eligibility. The full text of all potentially eligible studies was reviewed.

Assessment of risk of bias

We assessed risk of bias in included intervention studies with a modified version of the Cochrane Collaboration risk of bias tool, which assessed sequence generation and allocation concealment (selection bias), blinding of outcome assessors and validity of the outcome assessment tool (assessment bias), and

incomplete outcome data (attrition bias; considered high where <70% of randomised participants had outcome data available). We modified the tool by including an assessment of conflict of interest, judged as low when there was no evidence of industry involvement in study design, analysis, interpretation, or publication and no evidence that study authors received remuneration from relevant industry partners for other activities.

Data extraction

DI, VG-L, RJB, and JL-B developed, piloted, and refined an Excel data extraction form. Two researchers (RJB, and TK or TA) extracted data in duplicate. Disagreements and uncertainties about data coding were discussed within the team with leads as follows: RJB (clinical queries), VG-L (dietetic queries), DI (analysis and coding queries), and JL-B (study design and statistics queries). For foreign language studies, VG-L extracted data with a native speaker of the relevant language (see acknowledgements section). We extracted all relevant data from included studies, including data that we could not meta-analyse. Data were extracted either with raw frequencies or crude or adjusted effect estimates. We undertook random effect meta-analysis. When this was not possible or not appropriate we summarised study results in a narrative table.

Data selection for analysis

We extracted outcome data that adhered to the intention to treat principle in preference to data based on per protocol analyses. When studies included multiple intervention groups, we performed pairwise comparisons in which we split the number of events and no events in the unexposed/control group to prevent double counting. When studies reported data at multiple time points within one of our predefined age groupings, we extracted the most complete dataset available, beyond the intervention period (that is, from age 1 onwards). This is the dataset with the largest denominator or when the denominator is identical for multiple time points then the largest numerator (number of events) is used. When studies reported multiple assessments of the same outcome at the same time point, we selected clinical assessments in preference to serological assessments and skin prick in preference to assessment of specific IgE of allergic sensitisation. The GINI study¹²⁻¹⁵ used generalised estimating equations (GEE) to generate odds ratios in some of their publications. This represents the most complete data available from the study, so we selected this in preference to other GINI study data analyses. Raw data or risk ratios were not given in the generalised estimating equations reports. So for analyses that include such data from the GINI study we calculated odds ratios in place of risk ratios to be able to include the most complete GINI study data available in meta-analysis. For some meta-analyses generalised estimating equation data from the GINI trial were either not reported or could not be used because of multiple

intervention groups, and in these meta-analyses we used the most complete non-generalised estimating equation data available.

Data synthesis

We presented pooled results for binary outcomes from intervention studies as risk ratios calculated from the frequencies given in the study or as odds ratios when pooled data included those from a generalised estimating equation analysis and/or adjusted effect estimates. We pooled risk ratios using the Mantel-Haenszel method (with continuity correction of 0.5 in studies with zero cell frequencies) and odds ratios using the generic inverse variance method in the statistical programme R (R version 3.1.0, 2014, www.r-project.org). Pooled data for continuous outcomes measured with similar scales are presented as mean differences with 95% confidence intervals. Heterogeneity was quantified with I^2 and classified as low (<25%), moderate (5-50%), high (50-75%), or extreme (>75%). Data were not pooled when $I^2 \geq 80\%$. We undertook subgroup analyses for meta-analyses with more than five studies and assessed for publication bias (small study bias) using funnel plots and Egger's asymmetry test when there were 10 or more studies in a meta-analysis. We undertook subgroup analyses according to low versus unclear/high overall risk of bias; high versus normal/low risk of disease; quasi-randomised controlled trial versus randomised controlled trial; low versus unclear/high risk of conflict of interest; multifaceted versus not multifaceted intervention; and casein versus whey dominant hydrolysate.

Patient involvement

The findings of this study form part of the UK government's review of advice to the general public on infant feeding. No patients were involved in setting the research question or the outcome measures, nor were they involved in developing plans for design or implementation of the study. No patients were asked to advise on interpretation or writing up of results.

Results

Our original search identified 16 289 original titles. Screening of titles, abstracts, and full text yielded 52 studies (37 intervention trials) of hydrolysed formula, including over 19 000 participants. There were 28 randomised controlled trials, six quasi-randomised controlled trials, and three controlled clinical trials describing allergic or autoimmune outcomes. Appendix 2 shows full details of the search results (PRISMA flow chart), and characteristics of included studies are summarised in tables A and B in appendix 3. Twenty three studies used partially hydrolysed formula, and in at least 15 cases this was one specific formula (Nan HA/Good Start/Nidina HA/Beba HA, Nestlé, Vevey, Switzerland). Eighteen studies used extensively hydrolysed formula, and five studies (three for extensively, two for partially hydrolysed formula) used hydrolysed formula as part of a multifaceted intervention. In 30 of 37 studies infants were

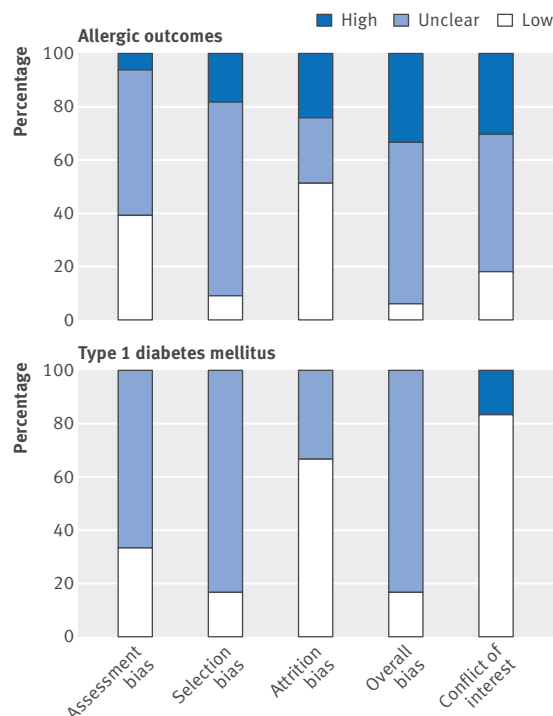


Fig 1 | Summary of risk of bias and conflict of interest in included studies reporting allergic outcomes and type 1 diabetes mellitus, showing proportion of studies with high, low, or unclear risk of bias in each domain

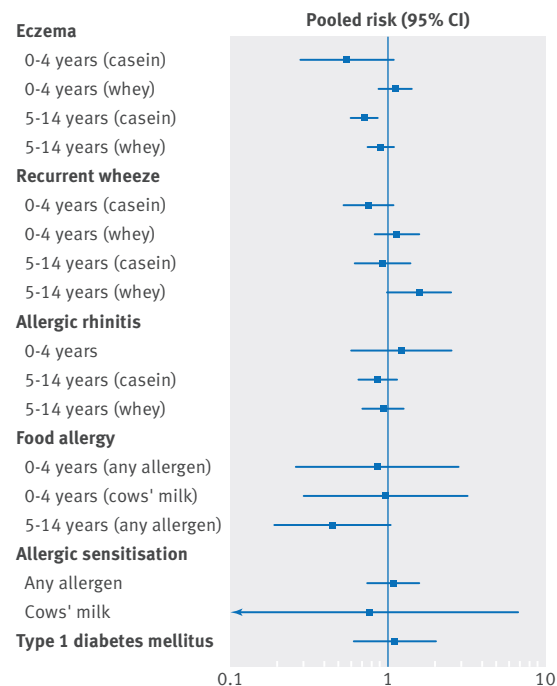


Fig 3 | Summary of treatment effects of hydrolysed formula on different outcome measures. Data shown are mean risk ratios (for allergic rhinitis at age 0-4; food allergy; allergic sensitisation; diabetes) or odds ratios (all other outcomes) with 95% confidence intervals for extensively hydrolysed formula compared with standard cows' milk formula

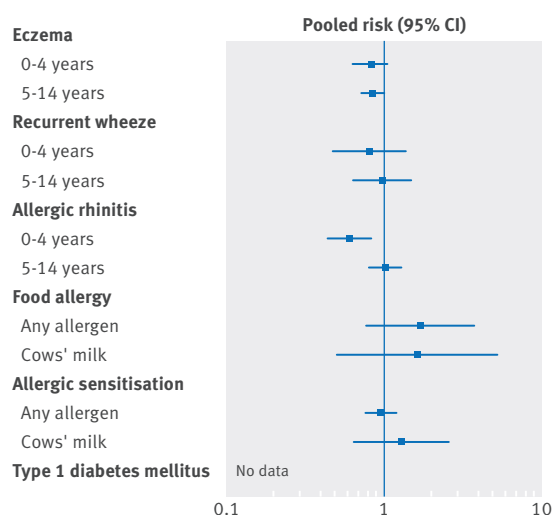


Fig 2 | Summary of treatment effects of hydrolysed formula on different outcome measures. Data shown are mean risk ratios (for allergic rhinitis at age 0-4; food allergy; allergic sensitisation; diabetes) or odds ratios (all other outcomes) with 95% confidence intervals for partially hydrolysed formula compared with standard cow's milk formula

at high risk of the relevant outcome(s) because of family history of disease in a first degree relative. Risk of bias and risk of conflict of interest were unclear or high in most studies for allergic outcomes (fig 1; table C in appendix 3), but low or unclear in most studies for autoimmune outcomes (fig 1; table D

in appendix 3). Figure 2 (partially hydrolysed formula) and figure 3 (extensively hydrolysed formula) summarise the findings of the main meta-analyses. Table 1 shows the GRADE assessment and summary of findings for key comparisons. Figures 4-7 show meta-analyses reported in table 1. The full report with a detailed description of all findings including meta-analyses and detailed methodology is available on the Food Standards Agency website (<http://www.food.gov.uk/science/research/allergy-research/fs305005>), together with an associated statement by the Committee on Toxicity (<http://cot.food.gov.uk/cotstatements>).

Risk of eczema

Twenty seven studies reported the effect of hydrolysed formula in infancy on risk of childhood eczema. The pooled data show no significant difference between extensively hydrolysed formula and standard cows' milk formula in risk of eczema at age 0-4 (odds ratio 0.84, 95% confidence interval 0.67 to 1.07; $I^2=30\%$) or age 5-14 (0.86, 0.72 to 1.02; $I^2=0\%$). Subgroup analysis suggested a significant difference in outcome according to study design or disease risk, with a more positive outcome in the single quasi-randomised controlled trial of normal risk infants. Analysis of data from randomised controlled trials for the most commonly studied partially hydrolysed formula (Nan HA/Good Start/Nidina HA/Beba HA, Nestlé, Vevey, Switzerland) showed no significant effect on risk of eczema at age 0-4 (0.94, 0.75

Table 1 | GRADE assessment and summary of key findings of review of studies of hydrolysed formula and risk of allergic or autoimmune disease

No of studies	Design	Risk of bias	Inconsistency	Indirectness	Imprecision	Publication bias	Other considerations	Odds ratio or relative risk	GRADE of evidence
Outcome: eczema at age 0-4									
Partially hydrolysed formula:									
12 studies	11 RCT, 1 qRCT	Serious. 11 studies with high or unclear overall risk of bias, all studies with high/unclear risk of conflict of interest	Not serious. $I^2=30\%$, study estimates varying from 0.33 to 1.44; subgroup analysis suggests difference by study design or population	No	Not serious. 95% CI for OR do not exclude clinically important effect, but exclude large effect sizes and significant harmful effects	No. NB Significant risk when pHF and eHF data combined. Egger's $P<0.05$	All RCTs undertaken in populations at high risk of eczema due to family history of allergic disease	OR 0.84 (0.67 to 1.07)	Moderate
Extensively hydrolysed formula:									
6 studies, 7 interventions	6 RCT	Serious. 5 studies with high or unclear overall risk of bias, all studies with high/unclear risk of conflict of interest	Serious. $I^2=74\%$ for analysis of casein eHF; 0% for whey eHF. Study estimates varying from 0.18 to 1.26	No	Serious 95% CI for OR do not exclude large beneficial or harmful effects	Not tested ($n<10$) NB Significant risk when pHF and eHF data combined. Egger's $P<0.05$	All RCTs undertaken in populations at high risk of eczema due to family history of allergic disease	Casein eHF OR 0.55 (0.28 to 1.09), whey eHF OR 1.12 (0.88 to 1.42)	Very low
Outcome: recurrent wheeze at age 0-4									
Partially hydrolysed formula:									
5 studies	5 RCT	Serious. 4 studies with high or unclear overall risk of bias, all studies with high/unclear risk of conflict of interest	No. $I^2=15\%$, study estimates varying from 0.29 to 1.20	No	Not serious. 95% CI for OR do not exclude clinically important effect, but exclude large effect sizes	Not tested ($n<10$) NB Significant risk when pHF and eHF data are combined. Egger's $P<0.05$	All RCTs undertaken in populations at high risk of allergy due to family history of allergic disease	OR 0.82 (0.48 to 1.41)	Moderate
Extensively hydrolysed formula:									
5 studies, 6 interventions	5 RCT	Serious. 5 studies with high or unclear overall risk of bias, all studies with high/unclear risk of conflict of interest	Serious. $I^2=74\%$ for analysis of casein eHF; 0% for whey eHF. Study estimates varying from 0.18 to 1.26	Not serious. 2 studies used multifaceted interventions	Not serious. 95% CI for OR do not exclude clinically important effect, but exclude large effect sizes	Not tested ($n<10$) NB Significant risk when pHF and eHF data combined. Egger's $P<0.05$	All RCTs undertaken in populations at high risk of allergy due to family history of allergic disease	Casein eHF OR 0.76 (0.53 to 1.09), whey eHF OR 1.15 (0.84 to 1.59)	Very low
Outcome: allergic sensitisation to cows' milk at any age									
Partially hydrolysed formula:									
7 studies	7 RCT	Serious. 6 studies with high or unclear overall risk of bias, and high/unclear risk of conflict of interest	No. $I^2=0\%$, study estimates varying from 0.44 to 9.63	Not serious. 2 studies used multifaceted interventions	Not serious. 95% CI for RR do not exclude clinically important effect, but exclude very large effect sizes	Not tested ($n<10$)	All RCTs undertaken in populations at high risk of allergy due to family history of allergic disease	RR 1.30 (0.65 to 2.60)	Moderate
Extensively hydrolysed formula:									
3 studies	3 RCT	Serious. All studies with high or unclear overall risk of bias, 2 studies with high/unclear risk of conflict of interest	Serious. $I^2=77\%$, study estimates varying from 0.08 to 10.13	Not serious. 1 study used multifaceted intervention	Serious. 95% CI for RR do not exclude large effect sizes	Not tested ($n<10$)	All RCTs undertaken in populations at high risk of allergy due to family history of allergic disease	RR 0.77 (0.09 to 6.73)	Very low
Outcome: type 1 diabetes mellitus at any age									
Extensively hydrolysed formula:									
5 studies	5 RCT	Not serious. All studies had low or unclear overall risk of bias; 4 studies had low risk of conflict of interest	Not serious. $I^2=25\%$, study estimates varying from 0.62 to 2.02	No	Not serious. 95% CI for RR do not exclude clinically important effect, but exclude large effect sizes	Not tested ($n<10$)	All RCTs undertaken in populations at high genetic risk of T1DM, and 4 of 5 studies used casein eHF	RR 1.12 (0.62 to 2.02)	High

RCT=randomised controlled trial, qRCT=quasi-randomised controlled trial, pHF=partially hydrolysed formula, eHF=extensively hydrolysed formula, RR=risk ratio, OR=odds ratio.

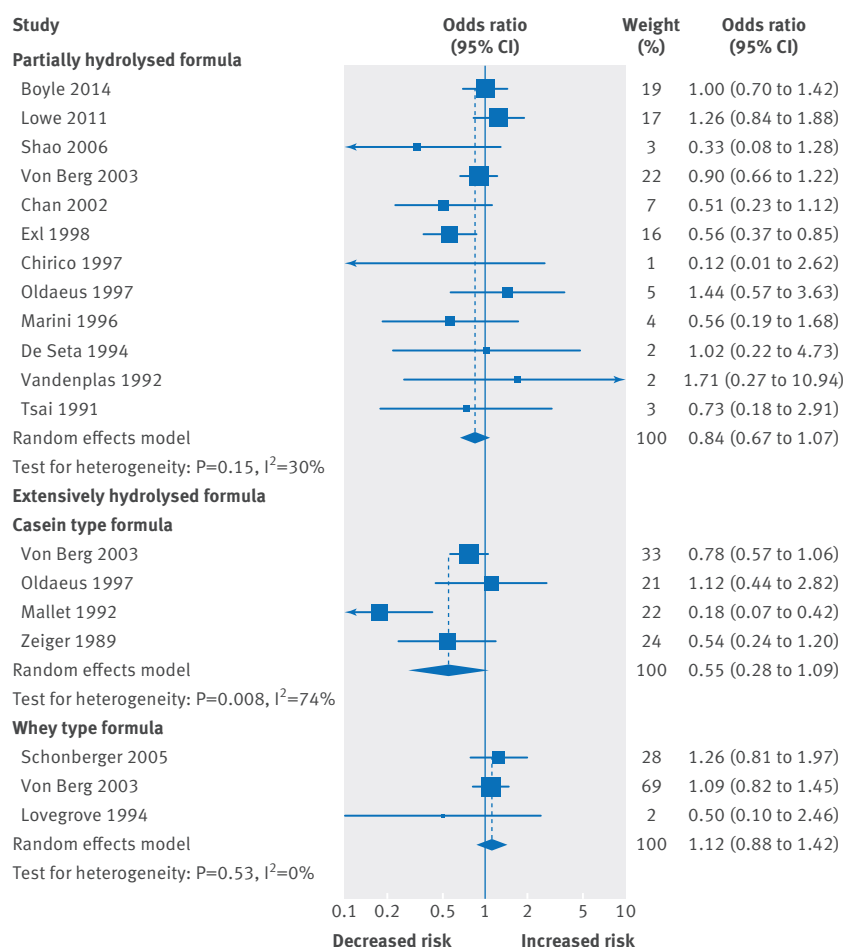


Fig 4 | Evidence from randomised or quasi-randomised controlled trials for partially hydrolysed formula and extensively hydrolysed formula compared with standard cows' milk formula for prevention of eczema in children aged ≤ 4

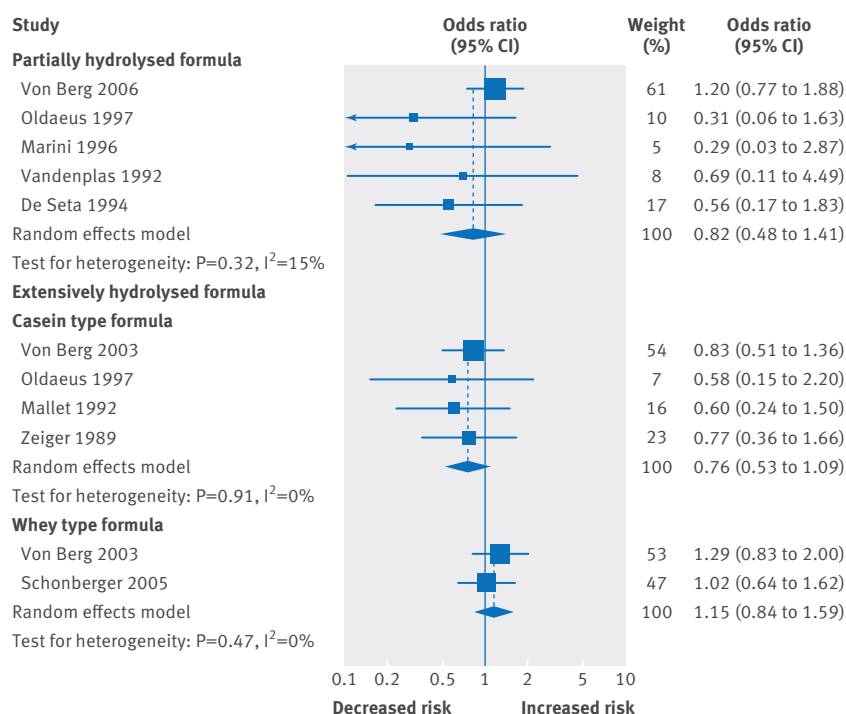


Fig 5 | Evidence from randomised controlled trials for partially hydrolysed formula and extensively hydrolysed formula compared with standard cow's milk formula, and recurrent wheezing at age ≤ 4

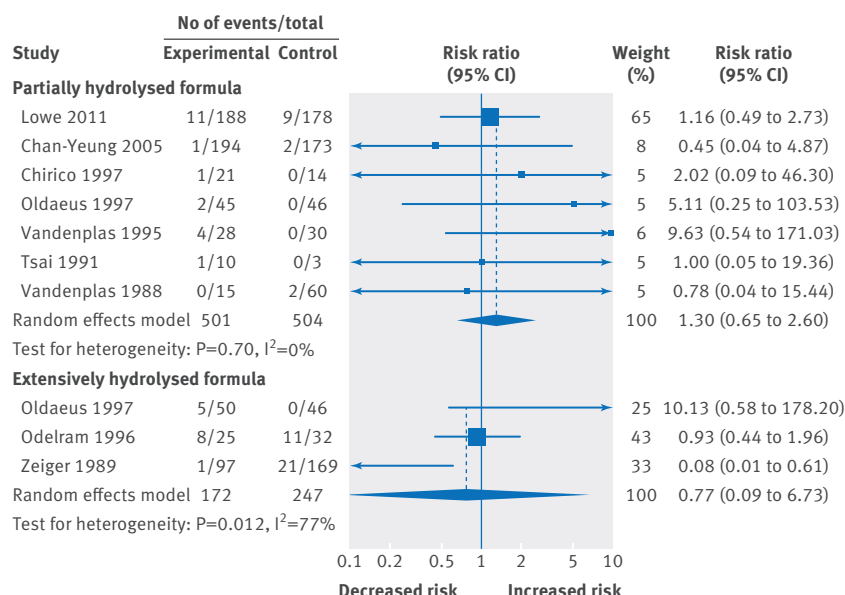


Fig 6 | Evidence from randomised controlled trials for partially hydrolysed formula and extensively hydrolysed formula compared with standard cow's milk formula, and allergic sensitisation to cows' milk

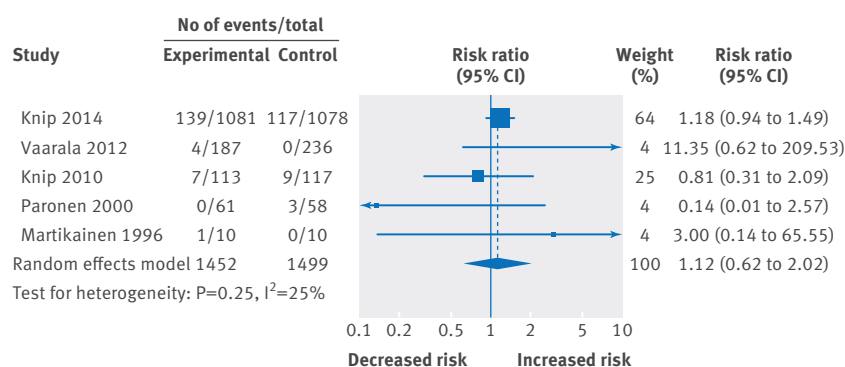


Fig 7 | Evidence from randomised controlled trials for extensively hydrolysed formula compared with standard cows' milk formula, and type 1 diabetes mellitus

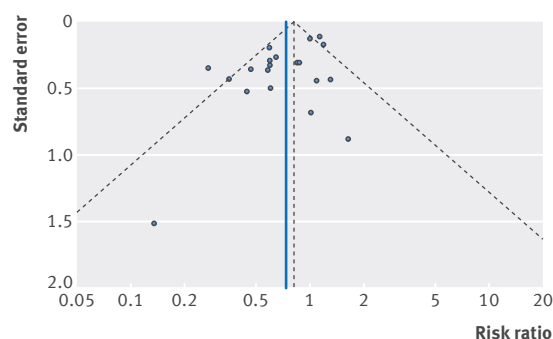


Fig 8 | Funnel plots for pooled analysis of "any hydrolysed formula" and risk of eczema showing evidence of publication bias/small study effects at age ≤ 4 (Egger's test $P=0.019$)

to 1.17; $I^2=0\%$). Though a meta-analysis of two studies on extensively hydrolysed formula found reduced odds of eczema at age 5-14 in children fed casein dominant formula during infancy (0.71, 0.59 to 0.87; $I^2=0\%$), this

effect was not seen for eczema at age 0-4 or for whey dominant formula at any age (0-4 or 5-14). Direct comparison of extensively versus partially hydrolysed formula, and casein versus whey dominant extensively hydrolysed formula, did not show a significant difference in risk of eczema at age 0-4 or 5-14. When we combined data for partially and extensively hydrolysed formula as "any hydrolysed formula" there was evidence of reduced eczema at age 0-4 (but not at age 5-14). This analysis necessitated use of per protocol data from the GINI study, had high statistical heterogeneity, and showed evidence of publication bias (fig 8).

Risk of wheeze

Twenty one studies reported the effect of hydrolysed formula in infancy on risk of wheeze or recurrent wheeze. Analysis of the outcome "wheeze" was inconclusive, with meta-analyses of partially hydrolysed formula dominated by a multifaceted intervention study in which uptake of the intervention was low and a quasi-randomised controlled trial with high risk of bias and conflict of interest. Meta-analysis of extensively hydrolysed formula was not possible because of extreme heterogeneity. Analysis of the outcome "recurrent wheeze" showed no significant difference between partially hydrolysed formula and standard cows' milk formula for recurrent wheeze at age 0-4 (odds ratio 0.82, 95% confidence interval 0.48 to 1.41; $I^2=15\%$) or age 5-14 (0.99, 0.65 to 1.51; $I^2=43\%$), and findings were also not significant for meta-analysis of casein or whey dominant extensively hydrolysed formula at ages 0-4 or 5-14. Direct comparison of extensively versus partially hydrolysed formula, and casein versus whey dominant extensively hydrolysed formula, did not show a significant difference in risk of recurrent wheeze at age 0-4 or 5-14. When data for both types of formula were combined as "any hydrolysed formula" there was no evidence of reduced recurrent wheeze at age 0-4 or age 5-14—this analysis necessitated use of per protocol data for the GINI study, and showed evidence of publication bias (fig 9).

Risk of allergic rhinitis

Twelve studies reported the effect of hydrolysed formula on risk of allergic rhinitis. At age 0-4 partially

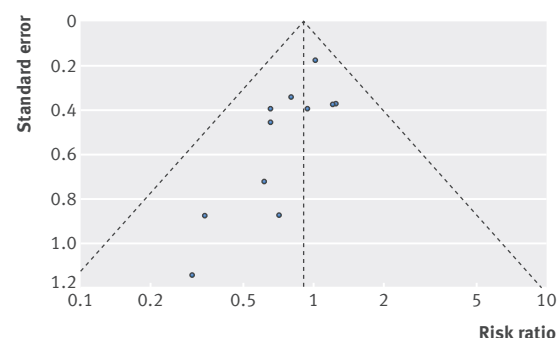


Fig 9 | Funnel plots for pooled analysis of "any hydrolysed formula" and recurrent wheeze showing evidence of publication bias/small study effects at age ≤ 4 (Egger's test $P=0.021$)

hydrolysed formula (but not extensively hydrolysed formula) was associated with significantly reduced risk of allergic rhinitis, though this meta-analysis was dominated by a multifaceted intervention study in which uptake of the intervention was low. At age 5-14 neither partially (odds ratio 1.03, 95% confidence interval 0.82 to 1.30; $I^2=0\%$) nor extensively (casein 0.87, 0.66 to 1.15; $I^2=0\%$; whey 0.93, 0.69 to 1.26; single study) hydrolysed formula were associated with a significant reduction in allergic rhinitis. Direct comparison of the two formulas, and casein versus whey dominant extensively hydrolysed formula, did not show a significant difference in risk of allergic rhinitis at age 0-4 or 5-14.

Risk of food allergy or allergic sensitisation

Thirteen and 19 studies reported the effect of hydrolysed formula on risk of food allergy and allergic sensitisation, respectively. There was no significant difference in risk of "any food allergy" with partially (risk ratio 1.73, 95% confidence interval 0.79 to 3.80; $I^2=42\%$) or extensively (0.86, 0.26 to 2.82; $I^2=42\%$) hydrolysed formula compared with standard formula at age 0-4, nor for extensively hydrolysed formula at age 5-14. We also found no difference in food allergy to cows' milk, egg, or (partially hydrolysed formula only) peanut. Direct comparison of the two formulas (egg allergy) and casein versus whey dominant extensively hydrolysed formula showed no significant difference in risk of food allergy. There was no significant difference in risk of allergic sensitisation to cows' milk with partially (1.30, 0.65 to 2.60; $I^2=0\%$) or extensively (0.77, 0.09 to 6.73; $I^2=77\%$) hydrolysed formula, and no significant difference between groups for risk of allergic sensitisation to "any allergen" or raised total IgE level.

Risk of type 1 diabetes mellitus

Six studies reported the effect of extensively hydrolysed formula compared with standard formula in infancy on risk of type 1 diabetes mellitus. There was no significant difference in risk with extensively hydrolysed formula (risk ratio 1.12, 95% confidence interval 0.62 to 2.02; $I^2=25\%$). We did not identify any studies of partially hydrolysed formula and diabetes nor of hydrolysed formula and other autoimmune outcomes.

Generalisability and study conduct

In 18 of 33 (55%) included studies reporting allergic outcomes, there were features suggesting issues with generalisability and possibly with study conduct.¹⁶ In seven studies there was either exclusive formula feeding from birth in all randomised participants¹⁷⁻²² or total cessation of breastfeeding by 14 days.²³ In five studies, randomisation and allocation of formula milk occurred early: during pregnancy,²⁴ at birth in all^{25 26} or some²⁷ participants, or in the first four days after birth in most participants.²⁸ In one study, infants were enrolled on day one, and over half were exclusively formula fed,²⁹ and in another breast milk was purposefully withheld for the first three days of life in

some infants.³⁰ In two studies breastfeeding was not mentioned,^{31 32} and in another two studies^{33 34} it was unclear whether or not the study population was selected for a high rate of early formula feeding because of insufficient information in the report. The information presented in these studies suggests the possibility that more could have been done to promote the maintenance and revival of breastfeeding in the study populations, in accordance with the International Code of Marketing of Breast-milk Substitutes.¹⁶ In the 15 remaining studies of allergic disease^{12 35-48} and in all diabetes studies^{47 49-52} there was no evidence of adverse breastfeeding outcomes in study participants.

Discussion

Principal findings

In this systematic review of hydrolysed infant formula for reducing risk of allergic or autoimmune disease we found no consistent evidence to support a protective role for partially or extensively hydrolysed formula. Our findings conflict with current international guidelines, in which hydrolysed formula is widely recommended for young formula fed infants with a family history of allergic disease.⁵⁻¹⁰ Such recommendations might therefore need revision.

Comparison with other studies

In 1936, Grulee and Sanford suggested that formula feeding could be associated with eczema,³ and more recent studies suggest that infant exposure to intact cows' milk might increase the risk of type 1 diabetes mellitus due to immunological cross reactivity between human and bovine insulin.⁴ Allergenic peptides in milk are 10-40 kDa in size, and bovine insulin is 5.8 kDa; partially and extensively hydrolysed formulas are intended to have no peptides >5 kDa and ≥ 3 kDa, respectively.⁵³ By the avoidance of exposure to insulin or allergenic peptides in milk with partially or extensively hydrolysed formula, one might in theory prevent immunological sensitisation to insulin or milk and associated diseases such as type 1 diabetes, milk allergy, and eczema. We did not, however, find consistent evidence from intervention trials to support this.

The infant feeding guidelines that recommend hydrolysed formula for allergy prevention recognise that the evidence base for such a recommendation is weak and informed by analysis of a small number of trials with high risk of bias. In the current 2006 Cochrane review Osborn and colleagues concluded that there is limited evidence to support a role for hydrolysed formula in reducing allergy to cows' milk and no evidence for other specific allergic outcomes.^{9 10} This conclusion was based on a single study,²¹ in which the selected outcome is likely to be a poor measure of allergy to cows' milk as it was reported in over 40% of the control group, and such allergy has a prevalence of 2-7.5%.⁵⁴ In our current meta-analysis of three studies reporting allergy to cows' milk, including data from the same study that

defined allergy to cows' milk as challenge proved allergy with a positive allergy test result (3% prevalence in control group), we found no evidence to support the conclusion of the Cochrane review.

International guidelines also support the use of hydrolysed formula for high risk infants to prevent eczema.⁵⁻⁷ We found no consistent evidence to support this. We did find that studies of lower quality design (controlled clinical trials and quasi-randomised controlled trials) had more positive outcomes than truly randomised controlled trials, and we found evidence of publication bias, suggesting that there could be small unpublished trials showing increased risk of eczema or recurrent wheeze associated with hydrolysed formula. The conclusions of an independent FDA review, which supported a limited health claim that a whey based partially hydrolysed formula (Good-Start, Nestlé, Vevey, Switzerland) could reduce the risk of eczema in high risk infants, also do not seem to be justified.⁸ That approval was based largely on review of the findings of the GINI study, in which per protocol analysis rather than intention to treat analysis was used to inform the FDA decision as the intention to treat analysis had not yet been published.⁸ The health claim was also approved before publication of one trial of the same formula with significantly negative results.⁴² Our own meta-analysis of published randomised controlled trials of the same partially hydrolysed formula does not support the FDA approved health claim that there is evidence for use of a whey based partially hydrolysed formula from birth to 4 months for reducing risk of eczema in healthy infants who are not exclusively breastfed and have a family history of allergy.

Strengths and limitations of the study

Many studies of allergic outcomes included in this review had unclear or high risk of bias and evidence of conflict of interest, often because of inadequate methods of randomisation and treatment allocation (selection bias) and support of the study or investigators from manufacturers of hydrolysed formula. In many cases study participants were infants with early full formula feeding, so our findings might not be applicable to populations with more typical feeding patterns.

Concerns have been raised about possible research fraud in this topic of investigation. We did not include the implicated studies, which evaluated a partially hydrolysed formula (Nan HA/Good Start/Nidina HA/Beba HA, Nestlé, Vevey, Switzerland) for preventing allergic outcomes and were led by Chandra and colleagues.⁵⁵⁻⁵⁷ Data from the Chandra studies played an important role in a 2003 Cochrane review, which concluded that hydrolysed formula reduces allergic disease.⁵⁸ Subsequent systematic reviews have excluded the Chandra data. The evidence of publication bias that we found, however, suggests that these systematic reviews might still overestimate the possibility that hydrolysed formula has beneficial effects.

Conclusions and policy implications

Our analyses suggest that current recommendations to use hydrolysed formula in place of standard cows' milk formula to prevent allergy in infants at high risk should be revised. We found no consistent evidence to support the current recommendations and found evidence of publication bias, methodological biases, and conflict of interest in those studies reporting allergic outcomes. We suggest that any future trials on hydrolysed formula should be prospectively registered, independently funded, and include adequate oversight to ensure that they do not negatively impact on breastfeeding in study participants.

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Transparency declaration: RJB affirms that this manuscript is an honest, accurate, and transparent account of the study being reported; that no important aspects of the study have been omitted; and that any discrepancies from the study as planned and registered have been explained.

Data sharing: Dataset and statistical code are available from the Food Standards Agency at www.food.gov.uk

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Appendix 1: Search strategy

Appendix 2: PRISMA flowchart

Appendix 3: Supplementary tables A-D